



## SEQUENCE LISTING

<110> ASAKO, HIRONYUKI  
> MATSUMURA, KENJI  
> SHIMIZU, MASATOSHI  
> ITC, NOBUYA  
> WAKITA, RYUHEI

<120> PROCESS FOR PRODUCING OPTICALLY ACTIVE  
4-HALO-3-HYDROXYBUTANATE

<130> 12249

<140> US 10/004,115  
<141> 2001-12-06

<150> JP 2000-372704  
<151> 2000-12-07

<150> JP 2001-006144  
<151> 2001-01-15

<150> JP 2001-026594  
<151> 2001-02-02

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<151> 2001-06-11

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Tyr Thr Ala Val Thr Thr Ala Leu Lys Thr Gly Tyr Arg His Leu Asp  
35 40 45  
Cys Ala Trp Tyr Tyr Leu Asn Glu Gly Glu Val Gly Glu Gly Ile Arg  
50 55 60  
Asp Phe Leu Lys Glu Asn Pro Ser Val Lys Arg Glu Asp Ile Phe Val  
65 70 75 80  
Cys Thr Lys Val Trp Asn His Leu His Arg Tyr Glu Asp Val Leu Trp  
85 90 95  
Ser Ile Asp Asp Ser Leu Lys Arg Leu Gly Leu Asp Tyr Val Asp Met  
100 105 110  
Phe Leu Val His Trp Pro Ile Ala Ala Glu Lys Asn Gly Gln Gly Glu  
115 120 125  
Pro Lys Ile Gly Pro Asp Gly Lys Tyr Val Ile Leu Lys Asp Leu Thr  
130 135 140  
Glu Asn Pro Glu Pro Thr Trp Arg Ala Met Glu Lys Ile Tyr Glu Asp  
145 150 155 160  
Arg Lys Ala Arg Ser Ile Gly Val Ser Asn Trp Thr Ile Ala Asp Leu  
165 170 175  
Glu Lys Met Ser Lys Phe Ala Lys Val Met Pro His Ala Asn Gln Ile  
180 185 190

Glu Ile His Pro Phe Leu Pro Asn Glu Glu Leu Val Gln Tyr Cys Phe  
 185 210 205  
 Ser Lys Asn Ile Met Pro Val Ala Tyr Ser Pro Leu Gly Ser Gln Asn  
 210 215 221  
 Gln Val Pro Thr Thr Gly Glu Arg Val Ser Glu Asn Lys Thr Leu Asn  
 225 230 225 240  
 Glu Ile Ala Glu Lys Gly Gly Asn Thr Leu Ala Gln Val Leu Ile Ala  
 245 250 255  
 Trp Gly Leu Arg Arg Gly Tyr Val Val Leu Pro Lys Ser Ser Asn Pro  
 260 265 270  
 Lys Arg Ile Glu Ser Asn Phe Lys Ser Ile Glu Leu Ser Asp Ala Asp  
 275 280 285  
 Phe Glu Ala Ile Asn Ala Val Ala Lys Gly Arg His Phe Arg Phe Val  
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 Lys Asn Leu Ser Ala  
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Gly Val Gly Phe Gly Thr Phe Ala Ser Glu Gly Ser Lys Gly Glu Thr	
20 25 30	
tat act gct gtc acc act gcc ctg aag acc ggt tac cgt cac ttg gac	144
Tyr Thr Ala Val Thr Ala Leu Lys Thr Gly Tyr Arg His Leu Asp	
35 40 45	
tgt gcc tgg tac tac ctg aac gag ggt gag gtt ggt gag ggt atc cgt	192
Cys Ala Trp Tyr Tyr Leu Asn Glu Gly Glu Val Gly Glu Ile Arg	
50 55 60	
gac ttc ctg aag gag aac ccc tcg gtg aag cgt gag gac atc ttc gtc	240
Asp Phe Leu Lys Glu Asn Pro Ser Val Lys Arg Glu Asp Ile Phe Val	
65 70 75 80	
tgc acc aag gtg tgg aac cac ctc cac cgt tat gag gac gtc ctc tgg	288
Cys Thr Lys Val Trp Asn His Leu His Arg Tyr Glu Asp Val Leu Trp	
85 90 95	
tcc att gac gac tcc ctg aag cgt ctt gga ctt gac tac gtt gat atg	336
Ser Ile Asp Asp Ser Leu Lys Arg Leu Gly Leu Asp Tyr Val Asp Met	
100 105 110	
ttc ctc gtt cac tgg ccc att gct gcc gag aag aat ggc cag ggt gag	384
Phe Leu Val His Trp Pro Ile Ala Ala Glu Lys Asn Gly Gln Gly Glu	
115 120 125	
ccc aag att ggc cct gac ggc aaa tac gtc att ctc aag gac ctg acc	432
Pro Lys Ile Gly Pro Asp Gly Lys Tyr Val Ile Leu Lys Asp Leu Thr	

130

135

140

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cgc aag gcc agg tcc att ggt gtc tcc aac tgg aac att gcc gag ctt Arg Lys Ala Arg Ser Ile Gly Val Ser Asn Trp Thr Ile Ala Asp Leu 165 170 175	528
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gag att cac ccc ttc ctg ccc aac gag gag ctg gtg cag tac tgc ttc Glu Ile His Pro Phe Leu Pro Asn Glu Glu Leu Val Gln Tyr Cys Phe 195 200 205	624
tcc aag aac att atg ccc gtg gcc tac tct cct ctg ggc tcg cag aac Ser Lys Asn Ile Met Pro Val Ala Tyr Ser Pro Leu Gly Ser Gln Asn 210 215 220	672
cag gtt ccc acc acc ggt gag cgg gtc agc gag aac aag act ctg aac Gln Val Pro Thr Thr Gly Glu Arg Val Ser Glu Asn Lys Thr Leu Asn 225 230 235 240	720
gag atc gcc gag aag ggc ggc aac acc ctt gct cag gtt ctt att gcc Glu Ile Ala Glu Lys Gly Asn Thr Leu Ala Gln Val Leu Ile Ala 245 250 255	768
tgg ggt ctg cgc cgt ggc tac gtc gtt ctc ccc aag agc tcc aac ccc Trp Gly Leu Arg Arg Gly Tyr Val Val Leu Pro Lys Ser Ser Asn Pro 260 265 270	816
aag cgc att gag tcc aac ttc aag agc att gag ctc tcc gat gcc gac Lys Arg Ile Glu Ser Asn Phe Lys Ser Ile Glu Leu Ser Asp Ala Asp 275 280 285	864
ttt gaa gcc atc aat gcc gtt gcc aag ggt cgt cac ttc cgt ttc gtc Phe Glu Ala Ile Asn Ala Val Ala Lys Gly Arg His Phe Arg Phe Val 290 295 300	912
aac atg aag gat act ttc gga tat gat gtc tgg ccc gag gag acc gcc Asn Met Lys Asp Thr Phe Gly Tyr Asp Val Trp Pro Glu Glu Thr Ala 305 310 315 320	960
aag aac ctg tct gcg tga Lys Asn Leu Ser Ala *	978
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&lt;210&gt; 4

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<212> PRT  
<213> PENICILLIUM CITRINUM

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<210> 5  
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<213> PENICILLIUM CITRINUM

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OLIGONUCLEOTIDE PRIMER FOR PCR

<400> 8  
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<210> 9  
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<400> 9  
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<210> 11  
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<400> 11  
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<400> 14  
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 <213> ESCHERICHIA COLI

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 agtqaaggtt ccaaggcgaa aacctatnct qctgtcacca ctgcctgaa aaccggttac 180  
 cgtcncttgg actgtgcctg gtactacctg aacaagggtt aggttggta gggtnccgt 240  
 gacttcctga aggaaaaccc ctcggtgaag cgtgaggaca tcttcgtctg caccaagggt 300  
 tggaaccacc tccaccgtta tgaggacgtc ctctggtcca ttgacnactc cctgaagcgt 360  
 cttggacttg actacgttga tatgttccctc gttcaactggc ccattgtgtc cgaaaaaaaaat 420  
 ggccagggtt agccaaaaat tggccctgac gccaataacn tcnttctcaa ggacctgacc 480  
 gaaanccna ncccacctgg cgcgttatgg aaaaaatttn tgangatccc aaggccaggt 540  
 ccattggtgtt ttccaattgg accattgccc accttgagaa gatgtccaag ttngccaagg 600  
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<213> Artificial Sequence

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<212> DNA

<213> Artificial Sequence

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<211> 331

<212> DNA

<213> ESCHERICHIA COLI

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gcccgtaaaga ccgggttaccg	tcacttggac	tgtgccttgt	240
gttggtgagg gtatccgtga	actacactgaa	cgagggtgag	300
ttcgtctgca ccaagggtgtg	gagaaccctt	cggtgaagcg	331
caaccaccc	tgaggacatc		

<210> 19

<211> 743

<212> DNA

<213> ESCHERICHIA COLI

<400> 19

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cggtgagcgg	gtcagcgaga	acaagactct	gaacgagatc	gccggagaagg	180
ccttgctcag	gttcttattg	cctggggctt	gcccgtggc	tacgtcggtc	240
ctccaacccc	aagcgattt	agtccaaactt	caagagcatt	gagctctccg	300
tgaagccatc	aatgccgtt	ccaaagggtcg	tcacttccgt	atgcccactt	360
tttcggatat	gatgtctggc	ccgaggagac	cgccaagaaac	ttcgtcaaca	420
gaaattataa	aatnacaccn	acnaaaancc	aaagcganag	tgaaggatac	480
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caattcnccc	cttattnattt	aattcttttt	taanggggnc	aaattccncc	660
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<211> 21

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agaagaatgg ccagggttag cccaaagattt gcctgtacgg caaatacgtc attctcaagg 180  
acctgacccga gaaccccgag cccacatggc gcgttatggaa gaaggattttt gaggatcgca 240  
aggccaggtt cattgggtgtc tccaaacttggaa ccattggccga ctttgagaag atgtccaagt 300  
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## OLIGONUCLEOTIDE PRIMER FOR PCR

<400> 25  
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acc ggt tta gga aaa gca atg gcg att cgt ttt gcg aca gaa aaa gct 96  
 Thr Gly Leu Gly Lys Ala Met Ala Ile Arg Phe Ala Thr Glu Lys Ala  
 20 25 30

aaa gta gtt gtg aac tat cgt tcg aaa gaa gaa gct aac agc gtt 144  
 Lys Val Val Val Asn Tyr Arg Ser Lys Glu Glu Ala Asn Ser Val  
 35 40 45

tta gaa gaa att aaa aaa gtg ggc gga gag gct att gcc gtc aaa ggt 192  
 Leu Glu Glu Ile Lys Lys Val Gly Gly Glu Ala Ile Ala Val Lys Gly  
 50 55 60

gat gta aca gtt gag tct gat gtg atc aat tta gttcaa tct gct att 240  
 Asp Val Thr Val Glu Ser Asp Val Ile Asn Leu Val Gln Ser Ala Ile  
 65 70 75 80

aaa gaa ttt gga aag cta gac gtt atg att aat aac gca gga atg gaa 288  
 Lys Glu Phe Gly Lys Leu Asp Val Met Ile Asn Asn Ala Gly Met Glu  
 85 90 95

aat ccg gtt tcg tct cat gaa atg tct tta agt gat tgg aat aaa gtc 336  
 Asn Pro Val Ser Ser His Glu Met Ser Leu Ser Asp Trp Asn Lys Val  
 100 105 110

att gat acg aac tta acg gga gca ttt tta ggc agc cgt gaa gcg att 384  
 Ile Asp Thr Asn Leu Thr Gly Ala Phe Leu Gly Ser Arg Glu Ala Ile  
 115 120 125

aaa tat ttt gtg gaa aat gat att aag gga aca gtt att aac atg tcg 432  
 Lys Tyr Phe Val Glu Asn Asp Ile Lys Gly Thr Val Ile Asn Met Ser  
 130 135 140

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Ser	Lys	Sly	Gly	Met	Lys	Ile	Met	Thi	Glu	Thr	Leu	Ala	Ile	Gly	Tyr			
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gct	cca	aaa	ggt	att	cgt	gtt	aat	aac	att	gga	ccg	gga	gcg	att	aat		576	
Ala	Pro	Lys	Gly	Ile	Arg	Val	Asn	Asn	Ile	Gly	Pro	Gly	Ala	Ile	Asn			
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aca	ccg	att	aac	gtt	gag	aaa	ttt	gtt	gtt	gat	cct	gag	cag	cgt	gca	gat	624	
Thr	Pro	Ile	Asn	Ala	Glu	Lys	Phe	Ala	Asp	Pro	Glu	Gln	Arg	Ala	Asp			
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gta	gaa	agc	atg	att	cca	atg	gga	ttc	att	gga	gag	ccg	gaa	gaa	att		672	
Val	Glu	Ser	Met	Ile	Pro	Met	Gly	Tyr	Ile	Gly	Glu	Pro	Glu	Glu	Ile			
					210					215					220			
gca	gcg	gtt	gct	gca	tgg	cta	gct	tct	tca	gag	gca	agt	tat	gta	aca		720	
Ala	Ala	Val	Ala	Ala	Trp	Leu	Ala	Ser	Ser	Glu	Ala	Ser	Tyr	Val	Thr			
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245																240		
ggg	att	aca	ctc	ttt	gct	gac	ggc	ggt	atg	aca	cag	tac	cca	tca	ttc		768	
Gly	Ile	Thr	Leu	Phe	Ala	Asp	Gly	Gly	Met	Thr	Gln	Tyr	Pro	Ser	Phe			
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ggc	gtc	ggc	ttt	ggt	acc	ttc	gct	agt	gaa	ggt	tcc	aag	ggc	gag	acc		96	
Gly	Val	Gly	Phe	Gly	Thr	Phe	Ala	Ser	Glu	Gly	Ser	Lys	Gly	Glu	Thr			
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															30			
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Cys	Ala	Trp	Tyr	Tyr	Leu	Asn	Glu	Gly	Glu	Val	Gly	Glu	Gly	Ile	Arg			
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															60			
gac	ttc	ctg	aag	gag	aac	ccc	tgg	aag	cgt	gag	gac	atc	ttc	gtc		240		
Asp	Phe	Leu	Lys	Glu	Asn	Pro	Ser	Val	Lys	Arg	Glu	Asp	Ile	Phe	Val			
															65		70	
															75			
															80			

tgc acc aag gtg tgg aac cac ctc cac cgt tat gag gag gac gtc ctc tgg Cys Thr Lys Val Trp Asn His Leu His Arg Tyr Glu Asp Val Leu Trp 85 90 95	288
tcc att gag gag tcc ctg aag cgt ctt gga ctt gag tac gtt gat att Ser Ile Asp Asp Ser Leu Lys Arg Leu Gly Leu Asp Tyr Val Asp Met 100 105 110	336
tcc ctc gtt cac tgg ccc att gct gcc gag aag aat ggc cag ggt gag Phe Leu Val His Trp Pro Ile Ala Ala Glu Lys Asn Gly Gln Gly Glu 115 120 125	384
ccc aag att ggc cct gag ggc aaa tac gtc att ctc aag gac ctg acc Pro Lys Ile Gly Pro Asp Gly Lys Tyr Val Ile Leu Lys Asp Leu Thr 130 135 140	432
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gag aag atg tcc aag ttc gcc aag gtc atg cct cac gcc aac cag atc Glu Lys Met Ser Lys Phe Ala Lys Val Met Pro His Ala Asn Gln Ile 180 185 190	576
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tcc aag aac att atg ccc gtg gcc tac tct cct ctg ggc tcg cag aac Ser Lys Asn Ile Met Pro Val Ala Tyr Ser Pro Leu Gly Ser Gln Asn 210 215 220	672
cag gtt ccc acc acc ggt gag cgg gtc agc gag aac aag act ctg aac Gln Val Pro Thr Thr Gly Glu Arg Val Ser Glu Asn Lys Thr Leu Asn 225 230 235 240	720
gag atc gcc gag aag ggc ggc aac acc ctt gct cag gtt ctt att gcc Glu Ile Ala Glu Lys Gly Gly Asn Thr Leu Ala Gln Val Leu Ile Ala 245 250 255	768
tgg ggt ctg cgc cgt ggc tac gtc gtt ctc ccc aag agc tcc aac ccc Trp Gly Leu Arg Arg Gly Tyr Val Val Leu Pro Lys Ser Ser Asn Pro 260 265 270	816
aag cgc att gag tcc aac ttc aag agc att gag ctc tcc gat gcc gac Lys Arg Ile Glu Ser Asn Phe Lys Ser Ile Glu Leu Ser Asp Ala Asp 275 280 285	864
ttt gaa gcc atc aat gcc gtt gcc aag ggt cgt cac ttc cgt ttc gtc Phe Glu Ala Ile Asn Ala Val Ala Lys Gly Arg His Phe Arg Phe Val 290 295 300	912
aac atg aag gat act ttc gga tat gat gtc tgg ccc gag gag acc gcc Asn Met Lys Asp Thr Phe Gly Tyr Asp Val Trp Pro Glu Glu Thr Ala 305 310 315 320	960
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325	

<210> 29  
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 <212> DNA  
 <213> Artificial Sequence

<220>  
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 OLIGONUCLEOTIDE PRIMER FOR PCR

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 <213> Artificial Sequence

<220>  
 <223> DESCRIPTION OF ARTIFICIAL SEQUENCE DESIGNED  
 OLIGONUCLEOTIDE PRIMER FOR PCR

<400> 30  
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<210> 32  
 <211> 23  
 <212> DNA  
 <213> Artificial Sequence

<220>  
 <223> DESCRIPTION OF ARTIFICIAL SEQUENCE DESIGNED  
 OLIGONUCLEOTIDE PRIMER FOR PCR

<400> 32  
 cggatccgtt atccgcgtcc tgc 23

<210> 33  
 <211> 28  
 <212> DNA  
 <213> Artificial Sequence

<220>  
 <223> DESCRIPTION OF ARTIFICIAL SEQUENCE DESIGNED  
 OLIGONUCLEOTIDE PRIMER FOR PCR

<400> 33  
 cggatccgag cgcccaatac gcaaaccg 28

<210> 34  
 <211> 385  
 <212> PRT  
 <213> CORYNEBACTERIUM SP.

<400> 34  
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 1 5 10 15  
 Glu Ile Pro Lys Pro Glu Pro Gly Pro Gly Glu Val Leu Leu Glu Val  
 20 25 30  
 Thr Ala Ala Gly Val Cys His Ser Asp Asp Phe Ile Met Ser Leu Pro  
 35 40 45  
 Glu Glu Gln Tyr Thr Tyr Gly Leu Pro Leu Thr Leu Gly His Glu Gly

<210> 35  
<211> 1158  
<212> DNA  
<213> CORYNEBACTERIUM SP.

<220>  
<221> CDS  
<222> (1) ... (1158)

<400> 35

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atg aag gcg atc cag tac acg cga atc ggc gcg gaa ccc gaa ctc acg
Met Lys Ala Ile Gln Tyr Thr Arg Ile Gly Ala Glu Pro Glu Leu Thr
   1           5           10          15

```

48

gag att ccc aaa ccc gag ccc ggt cca ggt gaa gtg ctc ctg gaa gtc  
 Glu Ile Pro Lys Pro Glu Pro Gly Pro Gly Glu Val Leu Leu Glu Val  
 20 25 30

96

acc gct gct ggc gtc tgc cac tgg gac gac ttc atc atg agc ctg ccc

144

Thr Ala Ala Gly Val Cys His Ser Asp Asp Phe Ile Met Ser Leu Pro	35	40	45	
gaa gag cag tac acc tac ggc ctt ccc atc atc ggt gtc gac gaa ggc				192
Glu Glu Gln Tyr Thr Tyr Gly Leu Pro Leu Thr Leu Gly His Glu Glu	50	55	60	
gca ggc aag gtc gtc gtc gag ggt gtc gaa ggt ctc gac atc				240
Ala Gly Lys Val Ala Ala Val Gly Glu Val Glu Gly Leu Asp Ile	65	70	75	80
gga acc aat gtc gtc gtc tac ggg cct tgg ggt tgg ggc aac tgt tgg				288
Gly Thr Asn Val Val Tyr Gly Pro Trp Gly Cys Gly Asn Cys Trp	85	90	95	
cac tgc tca caa gga ctc gag aac tat tgc tct cgc gcc caa gaa ctc				336
His Cys Ser Gln Gly Leu Glu Asn Tyr Cys Ser Arg Ala Gln Glu Leu	100	105	110	
gga atc aat cct ccc ggt stc ggt gca ccc ggc gcg ttg gcc gag ttc				384
Gly Ile Asn Pro Pro Gly Leu Gly Ala Pro Gly Ala Leu Ala Glu Phe	115	120	125	
atg atc gtc gat tct cct cgc cac ctt gtc ccg atc ggt gac ctc gac				432
Met Ile Val Asp Ser Pro Arg His Leu Val Pro Ile Gly Asp Leu Asp	130	135	140	
ccg gtc aag acg gtg ccg ctg acc gac gcc ggt ctg acg ccg tat cac				480
Pro Val Lys Thr Val Pro Leu Thr Asp Ala Gly Leu Thr Pro Tyr His	145	150	155	160
gcg atc aag cgt tct ctg ccg aaa ctt cgc gga ggc tcg tac gcg gtt				528
Ala Ile Lys Arg Ser Leu Pro Lys Leu Arg Gly Gly Ser Tyr Ala Val	165	170	175	
gtc att ggt acc ggc ggt ctc ggc cac gtc gct att cag ctc ctc cgc				576
Val Ile Gly Thr Gly Leu Gly His Val Ala Ile Gln Leu Leu Arg	180	185	190	
cac ctc tcg gcg gca acg gtc atc gct ttg gac gtg agc gcg gac aag				624
His Leu Ser Ala Ala Thr Val Ile Ala Leu Asp Val Ser Ala Asp Lys	195	200	205	
ctc gaa ctg gca acc aag gta ggc gct cac gaa gtg gtt ctg tcc gac				672
Leu Glu Leu Ala Thr Lys Val Gly Ala His Glu Val Val Leu Ser Asp	210	215	220	
aag gac gcg gcc gag aac gtc cgc aag atc act gga agt caa ggc gcc				720
Lys Asp Ala Ala Glu Asn Val Arg Lys Ile Thr Gly Ser Gln Gly Ala	225	230	235	240
gca ttg gtt ctc gac ttc gtc ggc tac cag ccc acc atc gac acc gcg				768
Ala Leu Val Leu Asp Phe Val Gly Tyr Gln Pro Thr Ile Asp Thr Ala	245	250	255	
atg gct gtc gcc ggc gtc gga tca gac gtc acg atc gtc ggg atc ggg				816
Met Ala Val Ala Gly Val Gly Ser Asp Val Thr Ile Val Gly Ile Gly	260	265	270	
gac ggc cag gcc cac gcc aaa gtc ggg ttc ttc caa agt cct tac gag				864
Asp Gly Gln Ala His Ala Lys Val Gly Phe Phe Gln Ser Pro Tyr Glu	275	280	285	

.gct tgg gtc aca gtt ccc tat tgg ggt gcc cgc aac gag tgg atc gaa Ala Ser Val Thr Val Pro Tyr Trp Gly Ala Arg Asn Glu Leu Ile Gln	290	300	310	312
ttg att gag ctc gcc tcc gct ggt atc ttc gat atc ggt gat cgt gac Leu Ile Asp Leu Ala His Ala Ser Ile Thr Asp Ile Gly Gly Asp	305	311	315	320
ctt cag tct cga caa cgg tgc cga agc gta tcg acg act ggg tgg cgg Leu Gln Ser Arg Gln Arg Cys Arg Ser Val Ser Thr Thr Gly Cys Arg	325	330	335	340
aac gct cag cgg ccc tgc ggt tgg tct gta gta ccc aca Asn Ala Gln Arg Pro Cys Gly Cys Gly Pro Trp Ser Val Val Pro Thr	340	345	350	356
gcg gta gaa cga cag cgg aaa aac act gat gcc cgg ccc aat tcg att Ala Val Glu Arg Gln Arg Lys Asn Thr Asp Ala Arg Pro Asn Ser Ile	355	360	365	370
cgg ccg ggc atc agt gtc aga aat tcg gtg tgc gct agc tgc acg cct Arg Pro Gly Ile Ser Val Arg Asn Ser Val Cys Ala Ser Cys Thr Pro	370	375	380	382
cga tga Arg * 385				1158